

2021-01-31

Phytochemical analysis and in vitro antifungal evaluation of *Jatropha curcas* against Late Leaf Spot disease on groundnut

Francis, Magreth

Journal of Animal & Plant Sciences

<https://dspace.nm-aist.ac.tz/handle/20.500.12479/1159>

Provided with love from The Nelson Mandela African Institution of Science and Technology

Phytochemical analysis and *in vitro* antifungal evaluation of *Jatropha curcas* against Late Leaf Spot disease on groundnut

Magreth Francis^{1,2,3}, Musa Chacha¹, Patrick A. Ndakidemi¹, Ernest R. Mbega^{1,2}

¹Department of Sustainable Agriculture, Biodiversity and Ecosystem Management, Nelson Mandela African Institution of Science and Technology (NM- AIST), P.O. Box 447, Arusha, Tanzania.

²Centre for Research, Agricultural advancement, Teaching Excellence and Sustainability in Food and Nutrition Security (CREATES-FNS), P.O. Box 447, Arusha, Tanzania.

³Deutscher Akademischer Austausch Dienst (DAAD), German Academic Exchange Service

Corresponding author email: francism@nm-aist.ac.tz

Key words: GC MS, biological control, *Phaeosariopsis personata*, mycelial inhibition

Publication date 31/01/2021, <http://m.elewa.org/Journals/about-japs/>

1 ABSTRACT

This study was done to evaluate the antifungal efficacy of *Jatropha curcas* leaf extracts against groundnut late leaf spot disease caused by *Phaeosariopsis personata* (*P. personata*) and identify their bioactive compounds responsible for antifungal effects. *Jatropha curcas* leaves extracted sequentially through chloroform, ethyl acetate and methanol solvents were evaluated against the mycelial growth of *P. personata* by food poisoning method. About 0.1, 0.25 or 0.5 mg/ml (plant extract/water) of each extract were mixed in molten PDA poured into Petri dishes. Thereafter solidified amended PDA with extracts was kept at room temperature for 24 hours. A seven-day-old fungal plug (4mm diameter) of *P. personata* was plated at the middle of the Petri dishes in triplicates. Inoculation on PDA plates amended with fungicide Chlorothalonil (720g/L) or water was included as positive and negative control respectively. The results proved that *J. curcas* leaf extracts possessed fungicidal properties since they inhibited the growth of *P. personata*. Moreover the antifungal effect of *J. curcas* leaf extracts increased as concentration increased. Moreover, *J. curcas* leaf extracts highly inhibited mycelial growth by (85.78%) similar to standard fungicide (chlorothalonil) (88.37%) in this experiment. The presence of important compounds found in *J. curcas* leaf extracts by GC-MS supported their ability against *P. personata* pathogen. Among the major compounds identified with antifungal activity were hexadecanoic acid methyl ester, hexadecanoic acid ethyl ester, hexadecane, n-hexadecanoic acid, octadecanoic acid ethyl ester, phytol and 9, 12-octadecadienoic acid (Z,Z)-methyl ester. The potentiality of *J. curcas* extracts in managing groundnut late leaf spot disease was confirmed by their ability to inhibit the growth of *P. personata* and possession of important phytochemical compounds.

2 INTRODUCTION

Groundnut late leaf spot disease (LLS) caused by *Phaeosariopsis personata* (*P. personata*) is a major limiting factor to groundnut productivity in Tropics and Subtropics (Khedikar *et al.*, 2010). LLS disease causes a considerable damage in the groundnut production leading to severe leaf defoliation hence reduces both pods yields and

haulm by 23-47% (McDonald *et al.*, 1985; Waliyar *et al.*, 2000). Much efforts of managing this plant fungal pathogen have been developed. Fungicides application has remained as a primary strategy in managing plant diseases. Fungicides seem to be effective financially and manage fungal diseases immediately despite their

shortcomings causing pathogen disease resistant and detrimental effects to human and environment (Karaman *et al.*, 2003; Monyo *et al.*, 2009). Application of the natural bioactive compounds originated from plant resources has gained much interest aiming to replace the synthetic compounds (Karaman *et al.*, 2003; Monyo *et al.*, 2009). This interest is based on possession of phytochemicals, which act differently against pathogens (Sharstry *et al.*, 2010; Gurjar *et al.*, 2012). *Jatropha curcas* (*J. curcas*) is cultivated in subtropical and semiarid regions, mainly as potential source traditionally used for medicinal purposes (Fairless, 2007). Moreover *J. curcas* extracts from various parts i.e.

leaves, stem, barks, roots, seed and seed oil have shown antifungal properties (Saetae and Suntornsuk, 2010). According to Siva (2008), *J. curcas* among 20 plant species was proved to have fungicidal property. Also according to Rahman *et al.* (2011), *J. curcas* fruit was reported its antifungal property. Furthermore, *J. curcas* leaf extract reported to inhibit the growth of *C. musae* causing anthracnose disease in banana. These few evidences suggest the fungicidal property of *J. curcas*. The study assessed the effectiveness of *J. curcas* leaf extracts against LLS that causes severe groundnut yield losses also identify the phytochemical compounds responsible for the management of named pathogen.

3 MATERIAL AND METHODS

3.1 Plant leaves: Plant leaves samples (*J. curcas*) were obtained from different parts in Arusha, Tanzania. Thereafter were washed, air-dried and ground to powder for extraction. "Pendo" groundnut variety, which is popular and highly susceptible to LLS disease, was

obtained from Naliendele Agricultural Research Institute, Mtwara, Tanzania. Pendo variety is early maturity (90-100 days), has high yield performances, easy to harvest and pluck (Bucheyeki *et al.*, 2010).



Plate 1: Symptoms of late leaf spot caused by *Phaeoisariopsis personata* on groundnut leaves

3.2 Preparation of leaf extracts: One kilogram of *J. curcas* powdered leaf was separately and sequentially extracted through different solvents in order of polarity chloroform (non-polar), ethyl acetate (mid-polar) and lastly on methanol (polar) at room temperature. Thereafter the leaf extracts were filtered by using Whatman no. 1 thereafter concentrated using

rotatory evaporator to give a sticky semisolid extract, which was kept in the refrigerator at 4°C.

3.3 Isolation of pathogen and culture preparation : Groundnut leaves showing black and nearly circular spots appear on the lower side of the leaflet were obtained from the farmer's fields from Singida and Dodoma regions, Tanzania. The isolation of the intended pathogen was done in the laboratory by adopting

the scheduled technique (Riker and Riker, 1936). Where the diseased leaf portions were cut into small pieces (1-2mm) sterilized with 0.1% mercuric chloride solution by soaking for 5 minutes then rinsed thrice with sterile distilled water (SDW) and dried on blotter paper. Thereafter those small pieces of leaves were plated of Potato Dextrose Agar (PDA) in a laminar hood then incubated at a room temperature $28 \pm 2^\circ\text{C}$ for 7 days to allow fungi to grow. The emerged fungal colonies were sub cultured to a fresh PDA plates thereafter incubated at a room temperature for 7 days in order to obtain *P. personata* culture. Fungal pathogen *P. personata* was identified by a single spore method. Fungal mycelium from the fresh culture examined under Sterio-microscope (Magnification 40X) by observing their morphological and distinctive images/features (Agrios 2005).

3.4 In vitro test of *J. curcas* leaf extracts on *P. personata*: The antifungal activity of chloroform, ethyl acetate and methanol leaf extracts of *J. curcas* against *P. personata* was measured by using a food poisoning technique by adopting the technique described by Kritzinger *et al.* (2005) with some modification. The appropriate amounts of each stock of extract was added to 100 ml of PDA before pouring into Petri dishes to yield final concentrations of 0.1, 0.25 and 0.5 mg/ml. Plugs (5 ml diameter) of *P. personata* from 7-day-old fungal culture was placed at the centre of the Petri dishes containing PDA amended with either chloroform, ethyl acetate and methanol leaf extracts of *J. curcas* or *P. hystrophorus* leaf extracts. The plates without phytoextract served as negative control and plate along with synthetic fungicide Chlorothalonil (720g/L) served as positive control. Treatments were arranged in a complete randomized design (CRD) with three replications and were conducted twice. The inoculated petri plates were incubated at room temperature and the radial growth was recorded

when the fungus reached the edge of the petri plates. The Percent inhibition of mycelial growth was calculated by comparing with mycelial growth of treatments/extracts and control following a standard proposed formular by Sivakumar *et al.* (2000);

$$I = [C - T / C] \times 100$$

Where;

I = Percent inhibition, **C** = Colony diameter in control, **T** = Colony diameter in treatment

3.5 Phytochemical analysis: The phytochemical analysis of *J. curcas* extracts was done by using Gas chromatography and mass spectroscopy (GC MS) at Tropical Pesticides Research Institute (TPRI), Arusha-Tanzania. The analysis was done using 7890A GC connected to Agilent 5975 MSD (Agilent technology, USA). Helium was used as carrier gas at 1.2ml/min flow rate. The GC was equipped with capillary column (HP 5) length of 30 meters, film 0.25 μm and internal diameter 0.250mm and temperature limit 50°C to 340°C (360°C) was used. The initial oven temperature was 50°C for 2min and then increased by $10^\circ\text{C}/\text{min}$ rise in temperature (i.e. $50\text{--}280^\circ\text{C}$). The injection volume was $1\mu\text{l}$ at a concentration of 1mg/ml of each sample. The mass spectra ionization voltage was 70eV and the total time taken for the analysis was 35min. The inlet temperature was 250°C . Each peak in the chromatography was identified basing on the retention index and compared the fragmentation pattern of the compounds with the mass spectra in the National Institute Standard and Technology (NIST) library.

3.6 Statistical analysis : Data were subjected to 3-way ANOVA (analysis of variance) in factorial arrangement, using STATISTICA program. The treatment means were compared by applying Fischer's least significant difference (LSD) at 5% level of significance.

4 RESULTS

4.1 *In vitro* evaluation of *J. curcas*: The antifungal efficacy of *J. curcas* leaf extracts at three level concentrations (0.1, 0.25 and 0.5mg/ml) was determined by observing the mycelial growth of *P. personata*. The mycelial growth inhibition of *P. personata* differed significantly at ($P \leq 0.001$) under different treatments, solvents and concentrations. The treatments amended with chlorothalonil (standard fungicide) and *J. curcas* leaf extract inhibited *P. personata* mycelial growth highly (88.37%, 85.78%) respectively as compared with the negative control (untreated) (0.00%). Moreover methanolic leaf extracts *J. curcas* inhibited highly the mycelial growth (74.04%) followed by chloroform and ethyl acetate and (57.89%, 56.22%) respectively. Furthermore, the highest concentration of *J. curcas* leaf extracts

(0.5mg/ml) inhibited the *P. personata* mycelial growth highly (78.07%) as compared with the lowest concentration (0.1mg/ml) (54.33%) (Table 1).

4.2 Interactive Effects between Treatments, Solvents and Concentrations:

The mycelial growth of *P. personata* differed highly significantly under interaction of factors; i.e. Treatments and Solvents; and Treatments and Concentrations; (Table 1). Generally, *J. curcas* leaves extracted by methanol inhibited the mycelial growth *P. personata* compared to other solvents. Moreover, the mycelial growth of *P. personata* under different leaf extracts concentration differed significantly ($P \leq 0.001$) where high inhibition was experienced at the highest concentration compared with the lowest concentration.

Table 1: The influence of *J. curcas* extracts on mycelial growth of *P. personata*

Factors	Percent inhibition
Treatments	
<i>Jatropha curcas</i>	85.78±1.64 ^b
Positive control (chlorothalonil)	88.37±0.93 ^a
Negative control	0.00±0.00 ^c
Solvents	
Chloroform	56.22±7.90 ^b
Ethyl acetate	57.89±8.09 ^b
Methanol	74.04±8.37 ^a
Concentrations	
0.1 mg/ml	54.33±7.61 ^c
0.25 mg/ml	58.74±8.20 ^b
0.5 mg/ml	78.07±8.49 ^a
3-way ANOVA (F-value)	
Treatments	6761.46***
Solvents	9.78**
Concentrations	31.33***
Treatments *Solvents	13.21***
Treatments *Concentrations	12.80***
Solvents*Concentrations	0.21ns
Treatments *Solvents*Concentrations	0.77ns

Means with different letters indicate significant differences among treatments according to Fischer's least significant difference (LSD) test. *, **, ***: significant at ($P \leq 0.05$), $P \leq 0.01$, ($P \leq 0.001$) respectively, ns= not significant

4.3 Chemical Composition of Leaf Extracts: This study reveals that the use of organic solvents in extraction of selected plants

has identified different compounds by GC-MS. From chloroform leaf extracts of *J. curcas* the following important phytochemical compounds

were identified (Table 2), the major compounds were *n*-hexadecanoic acid (7.89%), phenol, 2,4-bis (1, 1-dimethylethyl) (4.04%), cyclotetracosane (1.23%), hexadecane (1.20%) and octacosane (1.02%). The following major phytochemical compounds were identified from ethyl acetate leaf extracts of *J. curcas*; phytol (9.31%), hexadecanoic acid ethyl ester (3.97%), phenol 2, 4-bis (1, 1-dimethylethyl) (3.37%) and 5-eicosene, (E) (2. 11%), (Table 3). The following phytochemical compounds were

identified from methanolic leaf extracts of *J. curcas*; phytol (26.75%), hexadecanoic acid methyl ester (14.32%), octadecanoic acid, methyl ester (2.79%), 9, 12-octadecadienoic (Z,Z)-methyl ester (2.33%) (Table 4). The detected phytochemical compounds with antifungal property from chloroform, ethyl acetate and methanolic leaf extracts of *J. curcas* with their retention times, peak area (%), molecular formula and formula are presented in Table 2, 3 and 4.

Table 2: Reported antifungal activity of phytochemical compounds obtained from *J. curcas* chloroform leaf extract

Retention time(min)	Compound name	Molecular formula	Molecular weight (g/mol)	References
10.629	dodecane, 2,6,11-trimethyl-	C ₁₅ H ₃₂	212.41	(Zhang <i>et al.</i> , 2015)
11.745	2-tetradecene, (E)-	C ₁₄ H ₂₈	196.37	(Shirani <i>et al.</i> , 2017)
11.905	tetradecane	C ₁₄ H ₃₀	198.39	(Begum <i>et al.</i> , 2016)
12.460	pentadecane	C ₁₈ H ₃₈	254.49	(Zhang <i>et al.</i> , 2015)
12.958	octacosane	C ₂₈ H ₅₈	394.76	(Zhang <i>et al.</i> , 2018)
13.192	sulfurous acid butyl decyl ester	C ₁₆ H ₃₄ O ₃ S	306.50	(Sharma, 2016)
13.267	heneicosane	C ₂₁ H ₄₄	296.57	(Ebrahimabadi <i>et al.</i> , 2016)
13.461	phenol 2,4-bis(1, 1-dimethylethyl)	C ₁₄ H ₂₂ O	206.32	(Manikandan <i>et al.</i> , 2017)
14.011	2-bromo dodecane	C ₁₂ H ₂₅ Br	249.23	(Manikandan <i>et al.</i> , 2017)
14.503	hexadecane	C ₁₆ H ₃₄	226.44	(Zhang <i>et al.</i> , 2015)
15.041	heptadecane, 9-octyl-	C ₂₅ H ₅₂	352.68	(Musa <i>et al.</i> , 2015)
15.401	heptacosane	C ₂₇ H ₅₆	380.73	(Bouzabata <i>et al.</i> , 2018)
16.002	2,4-dimethyldodecane	C ₁₄ H ₃₀	198.38	(Begum <i>et al.</i> , 2016)
16.488	pentadecane	C ₁₅ H ₃₂	212.41	(Yuan <i>et al.</i> , 2012)
17.009	ethanol, 2-(octadecyloxy)-	C ₂₀ H ₄₂ O ₂	314.50	(El-Din Mohy and Mohyeldin, 2018)
18.142	hentriacontane	C ₃₁ H ₆₄	436.84	(Ruban and Gajalakshmi, 2012)
18.457	geranylgeraniol	C ₂₀ H ₃₄ O	290.48	(Ashraf <i>et al.</i> , 2017)
18.542	octadecane	C ₁₈ H ₃₈	254.49	(Zhang <i>et al.</i> , 2018)
18.869	<i>n</i> -hexadecanoic acid	C ₁₆ H ₃₂ O ₂	256.42	(Omoruyi <i>et al.</i> , 2014)
19.584	12-methyl-E-E-2, 13-octadecadien-1-ol	C ₁₉ H ₃₆ O	280.00	(Vijayabaskar and Elango, 2018).
20.013	tetradecanal	C ₁₄ H ₂₈ O	212.37	(Passos <i>et al.</i> , 2003)
29.037	cyclotetracosane	C ₂₄ H ₄₈	336.64	(Buglio <i>et al.</i> , 2017)

Table 3: Reported antifungal activity of phytochemical compounds obtained from *J. curcas* ethyl acetate leaf extract by GC-MS

Retention time (min)	Compound name	Molecular formula	Molecular weight (g/mol)	References
7.539	1,2,3-ropanetriol, monoacetate	C ₅ H ₁₀ O ₄	134.13	(Teoh and Mashitah, 2012)
8.460	2,5-pyrrolidinedione	C ₈ H ₁₃ NO ₂	331.32	(Takayama <i>et al.</i> , 1982)
8.826	hexadecane	C ₁₆ H ₃₄	226.44	(Adeleye <i>et al.</i> , 2010); (Oliveira <i>et al.</i> , 2014)
9.273	methyl salicylate	C ₈ H ₈ O ₃	152.15	(Pawar and Thaker, 2006)
11.321	triacetin	C ₉ H ₁₄ O ₆	218.21	(Osuntokun and Olajubu, 2014)
11.813	heptadecane	C ₁₇ H ₃₆	240.5	(Zhang <i>et al.</i> , 2015)
11.899	8-hexadecenal, 14-methyl-, (Z)-	C ₁₇ H ₃₂ O	252.4	(Osuntokun and Olajubu, 2014)
12.952	undecane	C ₁₁ H ₂₄	156.31	(Wanxi <i>et al.</i> , 2013)
13.467	phenol, 2,4-bis(1,1-dimethylethyl)	C ₁₇ H ₃₀ OSi	278.50	(Jun <i>et al.</i> , 2018)
13.993	1-naphthalenol	C ₁₀ H ₈ O	144.17	(Kumar <i>et al.</i> , 2012)
14.337	2,6,10,14,18,22-tetracosahexaene	C ₂₄ H ₃₈	326.6	(Devakumar <i>et al.</i> , 2017)
15.658	heptadecane	C ₁₇ H ₃₆	240.48	(Zhang, <i>et al.</i> , 2015)
16.591	1H-indene, 1-ethylideneoctahydro-7a-methyl-	C ₁₂ H ₂₂	166.30	(Wang <i>et al.</i> , 2013)
16.889	E-14-hexadecenal	C ₁₆ H ₃₀ O	238.41	(Devakumar <i>et al.</i> , 2017)
17.106	1-tetradecene	C ₁₄ H ₂₈	196.37	(Tayung and Jha, 2014)
17.896	tetramethyl-2-hexadecen-1-ol	C ₂₀ H ₄₀ O	296.50	(El-Din Mohy and Mohyeldin, 2018)
18.868	n-hexadecanoic acid	C ₁₆ H ₃₂ O ₂	256.42	(Tyagi and Agarwal, 2017)
18.983	9,12-octadecadienoic acid (Z,Z)-	C ₁₉ H ₃₄ O ₂	280.40	(El-Din Mohy and Mohyeldin, 2018)
19.109	5-eicosene, (E)-	C ₂₀ H ₄₀	280.50	(Adibe <i>et al.</i> , 2019)
19.172	hexadecanoic acid ethyl ester	C ₁₈ H ₃₆ O ₂	284.47	(El-Din Mohy and Mohyeldin, 2018)
19.338	2-methyl-Z,Z-3,13-octadecadienol	C ₁₉ H ₃₆ O	280.50	(Adibe <i>et al.</i> , 2019)
20.179	9,17-octadecadienal, (Z)-	C ₁₈ H ₃₂ O	264.40	(Adibe <i>et al.</i> , 2019)
20.413	phytol	C ₂₀ H ₄₀ O	296.54	(Pejin <i>et al.</i> , 2014)
21.008	9,12,15-octadecatrienoic acid ethyl ester, (Z,Z,Z)-	C ₂₀ H ₃₄ O ₂	306.48	(El-Din Mohy and Mohyeldin, 2018)
21.186	heptadecanoic acid ethyl ester	C ₁₉ H ₃₈ O ₂	298.50	(Bashir <i>et al.</i> , 2019)
23.869	eicosane	C ₂₀ H ₄₂	282.50	(El-Naggar <i>et al.</i> , 2017)

Table 4: Reported antifungal activity of phytochemical compounds obtained from methanolic leaf extract of *J. curcas* by GC-MS

Retention time (min)	Compound name	Molecular formula	Molecular weight (g/mol)	References
7.539	1,2,3-propanetriol monoacetate	C ₅ H ₁₀ O ₄	134.13	(Teoh and Mashitah, 2012)
9.273	methyl salicylate	C ₈ H ₈ O ₃	152.15	(Essien <i>et al.</i> , 2015)
10.549	2-undecanone	C ₁₁ H ₂₂ O	170.29	(Bisht and Chanotiya, 2011)
10.841	indole	C ₈ H ₇ N	117.15	(Sumiya <i>et al.</i> , 2017)
10.898	decanoic acid methyl ester	C ₁₁ H ₂₂ O ₂	186.29	(Belakhdar <i>et al.</i> , 2015)
11.121	2-methoxy-4-vinylphenol	C ₉ H ₁₀ O ₂	150.17	(Guo <i>et al.</i> , 2008)
11.287	tert-hexadecanethiol	C ₁₆ H ₃₄ S	258.50	(Yang <i>et al.</i> , 2016)
11.653	phenol, 2,6-dimethoxy-	C ₈ H ₁₀ O ₃	154.16	(Yang <i>et al.</i> , 2016)
11.813	tetradecane	C ₁₄ H ₃₀	198.39	(Begum <i>et al.</i> , 2016)
11.905	cyclotetradecane	C ₁₄ H ₂₈	196.37	(Afrouzan <i>et al.</i> , 2018)
11.991	pentanoic acid ethyl ester	C ₇ H ₁₄ O ₂	130.18	(Sumiya <i>et al.</i> , 2017)
12.248	2-propenoic acid 3-phenyl-, methyl ester	C ₁₀ H ₁₀ O ₂	162.18	(Umaiyaambigai <i>et al.</i> , 2017)
12.334	diphenyl ether	C ₁₂ H ₁₀	170.21	(Zhang <i>et al.</i> , 2018)
13.198	pentadecane	C ₁₅ H ₃₂	212.41	(Zhang <i>et al.</i> , 2015)
13.272	tridecane	C ₁₃ H ₂₈	184.36	(Yuan <i>et al.</i> , 2012)
14.503	hexadecane	C ₁₆ H ₃₄	226.44	(Oliveira <i>et al.</i> , 2014)
16.706	heptadecane	C ₁₇ H ₃₆	240.47	(Musa <i>et al.</i> , 2015)
16.797	17-pentatriacontene	C ₃₅ H ₇₀	490.93	(Zhang <i>et al.</i> , 2015)
16.889	1-nonadecene	C ₁₉ H ₃₈	266.50	(Asong <i>et al.</i> , 2019)
17.015	E-15-heptadecenal	C ₁₇ H ₃₂ O	252.43	(Begum <i>et al.</i> , 2016)
17.192	8-hexadecenal 14-methyl-,	C ₁₇ H ₃₂ O	252.40	(Aja <i>et al.</i> , 2014)
17.787	cyclopentadecane	C ₁₅ H ₃₀ O	210.40	(Nakashima <i>et al.</i> , 2014)
18.474	hexadecanoic acid methyl ester	C ₁₇ H ₃₄ O ₂	270.45	(Belakhdar <i>et al.</i> , 2015)
18.777	1-octadecene	C ₁₈ H ₃₆	252.48	(Omoruyi <i>et al.</i> , 2014)
18.868	2-methyl-Z, Z-3, 13-octadecadienol	C ₁₉ H ₃₆ O	280.49	(Phatangare <i>et al.</i> , 2017)
18.983	oleic acid	C ₁₈ H ₃₄ O ₂	282.46	(Adibe <i>et al.</i> , 2019)
19.486	9,17-octadecadienal, (Z)-	C ₁₈ H ₃₂ O	264.40	(Walters <i>et al.</i> , 2004)
19.836	2-methyl-Z,Z-3,13-octadecadienol	C ₁₉ H ₃₆ O	280.28	(Adibe <i>et al.</i> , 2019)
20.288	9, 12-octadecadienoic acid (Z,Z)-methyl ester	C ₁₉ H ₃₄ O ₂	294.47	(Adibe <i>et al.</i> , 2019)
20.413	phytol	C ₂₀ H ₄₀ O	296.0	(Chukwunonye <i>et al.</i> , 2015)
20.556	octadecanoic acid methyl ester	C ₁₉ H ₃₈ O ₂	298.50	(Hema <i>et al.</i> , 2011)
21.129	behenic alcohol	C ₂₂ H ₄₆ O	326.60	(Banaras <i>et al.</i> , 2017)
21.186	octadecanoic acid ethyl ester	C ₂₀ H ₄₀ O ₂	312.53	(Chandrasekaran <i>et al.</i> , 2011)
21.380	3,7,11,15-tetramethyl-2-hexadecen-1-ol	C ₂₀ H ₄₀ O	296.53	(El-Din Mohy and Mohyeldin, 2018)
				(El-Din Mohy and Mohyeldin, 2018)

22.096	9,17-octadecadienal, (Z)-	C ₁₈ H ₃₂ O	264.40	(Chukwunonye <i>et al.</i> , 2015)
23.875	eicosane	CH	282.50	(Shirani <i>et al.</i> , 2017)
24.241	docosanoic acid methyl ester	C ₂₃ H ₄₆ O ₂	354.61	(Aida <i>et al.</i> , 2017)

5 DISCUSSION

The effect of *J. curcas* leaf extracts of chloroform, ethyl acetate and methanolic against *P. personata* was more similar to the standard fungicide. This is attributed by their ability to produce toxins, which act on named pathogen by reducing disease development (Kagale *et al.*, 2004; Gupta *et al.*, 2008). This agrees with the findings by Muklesur *et al.* (2011) *J. curcas* leaf extract inhibited the mycelial growth *C. gloesporioides* by 50% on rubber tree. Moreover, the results obtained from *in vitro* trial found that the antifungal activity of *J. curcas* extracts varied with the type of solvent used for extractions. The results showed that polar solvent (methanol) gave greater antifungal effects on mycelial growth of *P. personata* as compared to intermediate and non-polar extract (ethyl acetate and chloroform) respectively. Possibly the polar compounds extracted through methanol had higher antifungal properties than polar compounds. This corresponds with the study done by Sharma *et al.* (2016), the methanolic fraction of *J. curcas* marked antifungal activities against four pathogenic fungus strains. Furthermore, correspond with the findings by (Krishnananda *et al.*, 2017) where *J. curcas* methanolic root extract shown antifungal activity up to 23.1% growth inhibition against *Rhizoctonia*. In addition, the study showed that the mycelial growth of *P. personata* was highly inhibited at highest concentration of *J. curcas* extracts than lowest concentration this shows that they are more fungitoxic at higher

concentrations. This study corresponds with the investigation by (Amah *et al.*, 2009) where *J. curcas* extract inhibited the growth of *F. oxysporum* by 54% inhibition at highest concentration (80 mg/ml) as compared with 10% inhibition at the lowest concentration 20mg/ml. Likewise according to Bajpai *et al.* (2012); disease severity was lowered as the concentration of plant extracts increased in all tests. Furthermore, the fungal growth was minimized as plant extract concentration increased (Goel and Sharma, 2013). GC-MS analysis was performed on *J. curcas* leaf extracts through chloroform, ethyl acetate and methanol as these exhibited antifungal activities. The major phytochemical compounds identified from this study were hexadecanoic acid ethyl ester, hexadecane, *n*-hexadecanoic acid, hexadecanoic acid methyl ester, octadecanoic acid ethyl ester, phytol and 9, 12-octadecadienoic acid (Z,Z)-methyl ester. Amongst hexadecanoic acid ethyl ester, hexadecanoic acid methyl ester-, octadecanoic acid ethyl ester, hexadecane, *n*-hexadecanoic acid, hexadecane, *n*-hexadecanoic acid, and 9, 12-octadecadienoic acid (Z,Z)-methyl ester are fatty acid with exceptional to phyto being diterpene alcohol. According to Hema *et al.* (2011); Belakhdar *et al.* (2015); (Chukwunonye *et al.* (2015); (Banaras *et al.* (2017) the identified compounds play a great role as antifungal agent. Normally, the fatty compounds absorb the fungus since it has lipophilic nature (Bassey *et al.*, 2013).

6 CONCLUSION

This study showed that *J. curcas* leaf extracts has antifungal effect against *P. personata* since they possess important bioactive compounds such as hexadecanoic acid methyl ester, hexadecanoic acid ethyl ester, octadecanoic acid ethyl ester,

hexadecane, *n*-hexadecanoic acid, phytol and 9, 12-octadecadienoic acid (Z,Z)-methyl ester. Hence *J. curcas* is an important agent for managing the groundnut late leaf spot disease aiming to improve groundnut production.

7 ACKNOWLEDGEMENTS

Authors are grateful to DAAD (German Academic Exchange Service) and CREATES-FN (World Bank Project) through Nelson

Mandela Institute of Science and Technology for funding and Tropical Pesticides Research Institute (TPRI) for technical support.

8 REFERENCES

- Adeleye IA, Daniels FV, and Omadime M: 2010. Characterization of volatile components of epa-ijebu: a native wonder cure recipe. *Journal of Pharmacology and Toxicology*, 6: 97-100
- Adibe MK, IbokAdeniyi-Akee MG, Mukaram A. and Ajala OE: 2019. Chemical compositions and antioxidant activity of leaf and stem essential oils of *Bryophyllum pinnatum* (lam.) Kurz. *GSC Biological and Pharmaceutical Sciences*, 9(2):57-64.
- Afrouzan H, Azar T, Sedigheh Z. and Ali E: 2018. Chemical Composition and Antimicrobial Activities of Iranian Propolis. *Iranian Biomedical Journal*, 22(1): 50–65
- Agrios GN: 2005. Plant Pathology. 5th Ed., Elsevier Academic Press, Amsterdam, New York, USA.
- Aida HS, Safaa AA. and Khoulood MB: 2016. GC-MS spectroscopic approach and antifungal potential of bioactive extracts produced by marine macro algae. *The Egyptian Journal of Aquatic Research* 42(3):289-299
- Aja PM, Nwachukwu N, Ibiam UA, Igwenyi IO, Offor CE. and Orji UO: 2014. Comparative Gas Chromatography-Mass Spectrometry (GC-MS) Analysis of Chemical Compounds of *Moringa oleifera* Leaves and Seeds from Abakaliki. *Nigeria Advances in Life Science and Technology* 24:
- Amah CP. and Aliero AA: 2009. Efficacy of aqueous extracts of some selected medicinal plants in the control of *Fusarium oxysporum*. *Biotropic Research International Journal*. 1:39-43.
- Ashraf SA, Al-Shammari E, Hussain T, Tajuddin S. and Panda BP: 2017. In-vitro antimicrobial activity and identification of bioactive components using GC-MS of commercially available essential oils in Saudi Arabia. *Journal of Food Science and Technology*, 54(12): 3948-3958. <https://doi.org/10.1007/s13197-017-2859-2>
- Asong JA, Ndhlovu PT, Khosana NS, Aremu AO. and Otang-Mbeng W: 2019. Medicinal Plants Used for Skin-Related Diseases among the Batswanas in Ngaka Modiri Molema District Municipality, South Africa. *S. Afr. J. Bot.* 3:56-62
- Bajpai VK, Baek KH, Kim SE, Han EJ, Kwak M, Oh K, Kim CJ, Kim S. and Choi GJ: 2012. In vivo Antifungal Activities of the Methanol Extracts of Invasive Plant Species Against Plant Pathogenic Fungi. *Plant Pathology Journal* 28(3): 317-321.
- Banaras S, Javaid A, Shoaib A. and Ahmed E: 2017. Antifungal activity of *Cirsium arvense* extracts against phytopathogenic fungus *Macrophomina phaseolina*. *Planta Daninha* v35:e017162738. Doi: 10.1590/S0100-83582017350100014
- Bashir S, Jabeen K, Iqbal S, Javed S, and Naeem A: 2019. *Lantana camara*: Phytochemical Analysis and Antifungal Prospective. *Planta Daninha*, 37,
- Bassey IN, Ogbemudia FO, Harold KO and Dung KE: 2013. Combined Antifungal Effects of Extracts of *Jatropha curcas* and *Chromolaena odorata* on Seed Borne Fungi of *Solanum gilo* Raddi. *Bull. Env. Pharmacol. Life Science* 2: 13-17
- Begum, F., I., Mohankumar, R., M. Jeevan and K. Ramani, 2016. GC-MS Analysis of Bio-active Molecules Derived from *Paracoccus pantotrophus* FMR19 and the Antimicrobial Activity Against Bacterial Pathogens and MDROs. *Indian journal of microbiology*, 56(4), 426–432. <https://doi.org/10.1007/s12088-016-0609-1>

- Belakhdar G, Benjouad E and Abdennebi H: 2015. Determination of some bioactive chemical constituents from *Thesium humile* Vahl. *Journal of Material Environmental Science* 6 (10): 2778-2783
- Bisht D. and Chanotiya CS: 2011. 2-Undecanone rich leaf essential oil from *Zanthoxylum armatum*. *Nat. Prod. Commun.*, 6(1): 111-114
- Bouzabata A, Mahomoodally F. and Tuberoso C: 2018. Ethnopharmacognosy of *Echinops spinosus* L. in North Africa: a mini review. *J. Complement. Med. Res*, 8:40-52.
- Bucheyeki TL, Shenkalwa ME, Mapunda T. and Matata WL: 2010. The groundnuts client oriented research in Tabora, Tanzania. *African Journal of Agricultural Research* 5(5): 356-362.
- Bughio SH, Muhammad QS, Shahabuddin M, Shaista B, Moina AM and Ayaz AM: 2017. Chemical composition of the essential oils from *Tamarix dioica* and determination of its antibacterial activity. *International Journal of Food Properties*, 20(3):2660–2667, DOI: 10.1080/10942912.2017.1387138
- Chandrasekaran M, Senthilkumar A. and Venkatesalu V: 2011. Antibacterial and antifungal efficacy of fatty acid methyl esters from the leaves of *Sesuvium portulacastrum* L. *European Review for Medical and Pharmacological Sciences*. 15: 775-780
- Chiteka, Z. A., T. A. Kucharek, D. A. Knauff, D. W. Gorbett and F. M. Shokes, 1988. Components of resistance to late leafspot in peanut. I. Levels and variability implications for selection. *Peanut Science*, 15 (1):25-30.
- Chukwunonye MO, Kelechi IN. and Marycolette NE: 2015. The Chemical Constituents and Bioactivity of the seed (Fruit) extracts of *Buchholzia Coriacea* Engler (Capparaceae). *Journal of Applied Science Environment Management* 19 (4): 795- 801
- Devakumar J, Keerthana V. and Sudha SS: 2017. Identification of bioactive compounds by gas chromatography-mass spectrometry analysis of *Syzygium jambos* (L.) collected from Western Ghats Region Coimbatore, Tamil Nadu. *Asian Journal of Pharmaceutical Clinical Research*, 10(1): 364-369
- Ebrahimabadi AH, Movahedpour MM, Batooli H, Ebrahimabadi EH, Mazoochi A. and Qamsari, MM: 2016. Volatile compounds analysis and antioxidant, antimicrobial and cytotoxic activities of *Mindium laevigatum*. *Iranian Journal of Basic Medical Sciences* 19(12): 1337-1344
- El-Naggar NE, El-Ahmady NE, Ashraf Abdal-Aziz AE, Mamdouh A. and Noura SN: 2017. *In vitro* activity, extraction, separation and structure elucidation of antibiotic produced by *Streptomyces anulatus* NEAE-94 active against multidrug-resistant *Staphylococcus aureus*. *Biotechnology and Biotechnological Equipment*, 31(2): 418–430, DOI: 10.1080/13102818.2016.1276412
- Essien EE., Newby JS, Walker TM, Setzer WN and Ekundayo O: 2015. Characterization and Antimicrobial Activity of Volatile Constituents from Fresh Fruits of *Alchornea cordifolia* and *Canthium subcordatum*. *Medicines* (Basel, Switzerland), 3(1):1. <https://doi.org/10.3390/medicines3010001>
- Fairless D: 2007. Bio-fuel: The little shrub that could-may be. *Nature*, 449: 652-655.
- Goel A. and Sharma K: 2013. Effect of *Euphorbia Pulcherrima* Leaf and Inflorescence Extract on Various Cytomorphological Parameters of *Aspergillus fumigatus*. *International Journal of Biological Life Science and Engineering* 7: 7-10.
- Guo L, Jin-zhong W, Ting H, Tong C, Khalid R and Lu-ping Q: 2008. Chemical Composition, Antifungal and Antitumor Properties of Ether Extracts of *Scapania*

- verrucosa* Heeg and its Endophytic Fungus *Chaetomium fusiforme*. *Molecules* (13):2114–2125; DOI: 10.3390/molecules13092114
- Gupta C, Garg AP, Uniyal RC. and Kumari A: 2008. Antimicrobial activity of some herbal oils against common food borne pathogens. *African Journal of Microbiology Research* 2:254-261.
- Gurjar MS, Ali S, Akhtar M. and Singh KS: 2012. Efficacy of plant extracts in plant disease management. *Agricultural Sciences*, 3(3): 425-433.
- Hema R, Kumaravel S. and Alagusundaram K: 2011. GC-MS Determination of Bioactive components of *Murraya koenigii*. *Journal of Am Sci.* 7:80-83.
- Jun M, Rui-Rui X, Yao L, Di-Feng R. and Jun L: 2018. Composition, Antimicrobial and Antioxidant Activity of Supercritical Fluid Extract of *Elsholtzia ciliate*. *Journal of Essential Oil Bearing Plants*, 21(2): 556-562, DOI: [10.1080/0972060X.2017.1409657](https://doi.org/10.1080/0972060X.2017.1409657)
- Kagale S, Marimuthu T, Thayumanavan B, Nandakumar R and Samiyappan R: 2004. Antimicrobial activity and induction of systemic resistance in rice by leaf extract of *Datura metel* against *Rhizoctonia solani* and *Xanthomonas oryzae* pv. *Krishnananda Krishnananda*. *Physiol. Mol. Plant Pathology* 65: 91-100.
- Karaman I, Sahin P, Gulluce M, Oguten H, Songul M. and Adiguzel A: 2003. Antimicrobial activity of aqueous and methanol extracts of *Juniperus oxycedrus* L. *J Ethnopharmacology*, 2837:1-5
- Khedikar Y, Gowda M, Sarvamangala C, Patgar K, Upadhyaya H. and Varshney R: 2010. A QTL study on late leaf spot and rust resistance in groundnut (*Arachis hypogaea* L.). *Theoretical Applied Genetics*, 121:971-984.
- Krishnananda PI, Amit GD, Dipika AP, Mahendra SD, Mangesh PM, Vaibhav CK. and Dhiraj R G: 2017. Bioassay guided fractionation of antifungal activity of *Jatropha curcas*. *Journal of Pharmacognosy and Phytochemistry*, 6(6): 2147-2154
- Kritzinger Q, Lall N. and Aveling TAS: 2005. Antimicrobial activity of cowpea (*Vigna unguiculata*) leaf extracts. *South African Journal of Botany*, 71: 45-48.
- Kumar S, Kumar P. and Sati N: 2012. Synthesis and Biological evaluation of Schiff bases and azetidinones of 1-naphtol. *Journal of Pharmacological Biopesticides*, 4: 246-249
- Manikandan G, Vimala RA, Divya C. and Ramasubbu R: 2017. GC-MS analysis of phytochemical constituents in the petroleum ether leaf extracts of *Milletia Peguensis*. *International Research Journal of Pharmacy*, 8(9): 455-458
- McDonald D, Subrahmanam P, Gibbons RW. and Smith DH: 1985. Early and late leaf spots groundnut. Patancheru, India: ICRISAT Information Bulletin, No. 21.
- Mohy El-Din SM and Mohyeldin MM: 2018. Component Analysis and Antifungal Activity of the Compounds Extracted from Four Brown Seaweeds with Different Solvents at Different Seasons. *Journal Ocean University of China* 17: 1178-1188. <https://doi.org/10.1007/s11802-018-3538-2>
- Monyo ES, Osiru MO, Kadyampakeni D, Mponda O, Chinyamunyamu B: 2009. Improving Food Security and Nutrition in Malawi and Tanzania through Research on Edible Legumes. Proceedings of Stakeholder Workshops on Groundnut Production in Malawi and Tanzania held 1-2 March and 13 April 2007, Lilongwe (Malawi) and Mtwara (Tanzania).
- Muklesur R, Siti HA, Mahmud TM. and Mohammad Z: 2011. Extraction of *Jatropha curcas* fruits for antifungal activity against *Colletitrichum gloeosporoides* of papaya. *African Journal of Biotechnology* 10 (48):9796-9799.
- Musa AM, Ibrahim MA, Aliyu AB, Abdullahi MS, Tajuddeen N, Ibrahim H. and Oyewale AO: 2015. Chemical

- composition and antimicrobial activity of hexane leaf extract of *Anisopus mannii* (Asclepiadaceae). *Journal of intercultural ethnopharmacology*, 4(2): 129-133.
<https://doi.org/10.5455/jice.20150106124652>
- Nakashima TN, Masato I, Junya O, Yoshiyuki K, Kenichiro N, Atsuko M, Aki I, Kazuhiko O, Kazuro S, Yoko T. and Satoshi O: 2014. Mangromicins A and B: structure and antitrypanosomal activity of two new cyclopentadecane compounds from *Lechevalieria aerocolonigenes* K10-0216. *The Journal of Antibiotics* 67: 253-260
- Oliveira GT, Jaqueline MS, Rosa LH, Siqueira EP, Johann S. and Lima LARS: 2014. *In vitro* antifungal activities of leaf extracts of *Lippia alba* (Verbenaceae) against clinically important yeast species. *Revista da Sociedade Brasileira de Medicina Tropical* 47(2): 247-250.
- Omoruyi BE, Afolayan AJ. and Bradle G: 2014. Chemical composition profiling and antifungal activity of the essential oil and plant extracts of *Mesembryanthemum edule* (L.) bolus leaves. *African journal of traditional, complementary, and alternative medicine: AJTCAM*, 11(4): 19-30.
<https://doi.org/10.4314/ajtcam.v11i4.4>
- Osuntokun OT. and Olajubu FA: 2014. Comparative study of phytochemical and proximate analysis of seven Nigerian medicinal plants. *Applied Science Research Journal* 2(1): 10-26.
- Passos XS, An CM, Juliana SP, Ana CF, Garcia FC. and Maria RS: 2003. Composition and Antifungal Activity of the Essential Oils of *Caryocar brasiliensis*. *Pharmaceutical Biology*, 41(5): 319-324
- Pawar VC. and Thaker VS: 2006. *In vitro* efficacy of oils against *Aspergillus niger*. *Mycosis*, 49: 316-323.
- Pejin B, Savica A, Sokovicb M, Glamoclijab J, Ciricb A. and Nikolicb M: 2014. Further *in vitro* evaluation of antiradical and antimicrobial activities of phytol. *Nat Prod Res* 28: 372-6.
- Phatangare ND, Deshmukh KK, Murade VD, Hase GJ. and Gaje TR: 2017. Isolation and Characterization of Phytol from *Justicia gendarussa* Burm. f.-An Anti-Inflammatory Compound. *International Journal of Pharmacognosy and Phytochemical Research* 9(6): 864-872
- Rahman M, Ahmad STMH, Mohamed M, Zaki M. and Rahman A: 2011. Extraction of *Jatropha curcas* fruits for antifungal activity against anthracnose (*Colletotrichum gloeosporioides*) of papaya. *African Journal of Biotechnology*, 10(48): 9796-9799
- Riker and Riker: 1936. Introduction to research on plant diseases. John S. Swift and Co. Inc., St. Louis, Chicago.
- Ruban P. and Gajalakshmi K: 2012. *In vitro* antibacterial activity of Hibiscus rosa-sinensis flower extract against human pathogens. *Asian Pacific Journal of Tropical Biomedicine*, 2(5): 399-403.
[https://doi.org/10.1016/S2221-1691\(12\)60064-1](https://doi.org/10.1016/S2221-1691(12)60064-1)
- Saetae D. and Worapot S: 2010. Antifungal Activities of Ethanolic Extract from *Jatropha curcas* Seed Cake. *Journal of Microbiology and Biotechnology*, 20: 319-324.
- Sharma S, Amita K. and Mamta S: 2016. Comparative GC-MS Analysis of Bioactive Compounds in Methanolic Extract of *Calotropis gigantea* (L) W.T. Aiton Leaf and Latex. *International Journal of Pharmacognosy and Phytochemical Research* 8(11): 1823-1827
- Sharstry RA, Biradar-Mahadevan KM. and Habbu PV: 2010. Isolation and Characterization of Secondary Metabolite from *Amorphophallus paeoniifolius* for Hepatoprotective activity. *Research Journal of Pharmaceutical, Biological and Chemical Sciences*, 1(4):429-437.
- Shirani M, Samimi A, Kalantari H, Madani M. and Kord ZA: 2017. Chemical composition and antifungal effect of hydroalcoholic extract of *Allium tripedale*

- (Tvautv.) against *Candida* species. *Current medical mycology*, 3(1): 6-12. <https://doi.org/10.18869/acadpub.cm.m.3.1.6>
- Siva N, Ganesan S, Banumathy N. and Muthuchelian BN: 2008. Antifungal effect of leaf extract of some medicinal plants against *Fusarium* causing wilt disease of *Solanum melogena* L. *Ethnobotanical Leaflets*, 12: 156-163
- Sivakumar, D., Wilson W.R.S., Wijesundera R.L.C., Marikar F.M.T. and Abeyesekere M: 2000. Antagonistic effect of *Trichoderma harzianum* on postharvest pathogen of rambutan (*Nephelium lappaceum*). *Phytoparasitica*, 28: 240-247.
- Sumiya T, Mai I, and Keimei O: 2017. Synthesis of Imidazole and Indole Hybrid Molecules and Antifungal Activity against Rice Blast. *International Journal of Chemical Engineering and Applications*, 8(3):
- Takayama C, Akira F, Osamu K and Yoshio H: 1982. Quantitative Structure-activity Relationships of Antifungal 1-(3, 5-Dichlorophenyl)-2,5-pyrrolidinediones and 3-(3,5-Dichlorophenyl)-2,4-oxazolidinedione. *Agric. Biol Chem.*, 46(2): 2755-2758
- Tayung K. and Jha DK: 2014. Endophytic fungi as potential sources of bioactive natural products: prospects and challenges. Indian Society of Mycology and Plant Pathology Scientific Publishers (India), Jodhpur. *Revised Plant Pathology* 6: 299-334.
- Teoh PY and Mashitah M. D: 2012. Screening of antifungal activities from genera *Trametes* against growth of selected wood-degrading fungi from Malaysia. *Australian Journal of Basic and Applied Sciences* 6(1): 79-85
- Tyagi T. and Agarwal M: 2017. GC-MS analysis of invasive aquatic weed, *Pista Stratiotes* L. and *Eichhornia Crassipes* (Mart.) Solms. *International Journal of Current Pharmaceutical Research*, 9(3):111.
- Umaiyaibigai D, Saravanakumar K. and Adaikala RG: 2017. Phytochemical Profile and Antifungal Activity of Leaves Methanol Extract from the *Pydrax dicoccos* (Gaertn) Teys. & Binn. Rubiaceae Family. *International Journal of Pharmacology, Phytochemistry and Ethnomedicine*. 7: 53-61
- Vijayabaskar G. and Elango V: 2018. Determination of phytochemicals in *Withania somnifera* and *Smilax china* using GC-MS technique. *Journal of Pharmacognosy and Phytochemistry* 7(6): 554-557
- Waliyar F, Adomou M. and Traore A: 2000. Rational use of fungicide applications to maximize peanut yield under foliar disease pressure in West Africa. *Plant Disease*, 84: 120-121.
- Walters D, Raynor L. and Mitchell A: 2004. Antifungal Activities of Four Fatty Acids against Plant Pathogenic Fungi. *Mycopathologia* 157: 87-90 <https://doi.org/10.1023/B:MYCO.0000012222.68156.2c>
- Wang C, Zhifang W, Qiao X, Li Z, Fengjua L, Mianhua C, Yurong W, Yufang H and Haiyan C: 2013. Antifungal activity of volatile organic compounds from *Streptomyces alboflavus* TD-1. *FEMS Microbiology Letters*, 341(1):45-51
- Wanxi P, Zhi L, Junbo C, Fangliang G. and Xiangwei Z: 2013. Biomedical Molecular Characteristics of YBSJ Extractives from *Illicium Verum* Fruit. *Biotechnology and Biotechnological Equipment*, 27(6): 4311-4316
- Yang J, Cheng-Hong Y, Ming-Tsai L, Zi-Jie G, Yuh-Wern W, and Li-Yeh C: 2016. Chemical Composition, Antioxidant, and Antibacterial Activity of Wood Vinegar from *Litchi chinensis*. Chemical Composition, Antioxidant, and Antibacterial Activity of Wood Vinegar from *Litchi chinensis*. *Molecules*, 21:1150; doi:10.3390/molecules21091150
- Yuan J, Raza W, Shen Q. and Huang Q: 2012. Antifungal activity of *Bacillus amyloliquefaciens* NJN-6 volatile compounds against *Fusarium oxysporum* f.

- sp. cubense. *Applied and environmental microbiology*, 78(16): 5942-5944.
- Zhang P, Li X, Yuan XL, Du YM, Wang BG, and Zhang ZF: 2018. Antifungal Prenylated Diphenyl Ethers from *Arthrrium arundinis*, an Endophytic Fungus Isolated from the Leaves of Tobacco (*Nicotiana tabacum* L.). *Molecules* (Basel, Switzerland), 23(12): 3179. <https://doi.org/10.3390/molecules23123179>
- Zhang X, Xia C. and Li C: 2015. Chemical composition and antifungal activity of the volatile oil from *Epichloë gansuensis*, endophyte-infected and non-infected *Achnatherum inebrians*. *Sci. China Life Sci.* 58: 512-514.